THESIS

NODULES ON LEGUMES

CHARLES E. TILLSON, '07

THE LIFE HISTORY OF NODULES UPON LEGUMES AND THE EFFECT OF A GROWING LEGUME UPON OTHER PLANTS OF GRASS FAMILY GROWING ADJACENT.

THESIS

PRESENTED TO THE DEPARTMENT OF AGRONOMY OF THE
NEW YORK STATE COLLEGE OF AGRICULTURE
CORNELL UNIVERSITY

for the

DEGREE OF BACHELOR OF SCIENCE OF AGRICULTURE

by

CHARLES BURRITT TILLSON

1907.

CONTENTS

INTRODUCTION

The purpose of this thesis is to determine , if possible, the life history of nodules upon legumes and the effect of a growing legume upon other plants of the grass family growing adjacent. In an attempt to solve these problems, I have supplemented my field work with a thorough study of some of the more important legumes in a green house.

The field work was commenced on Oct. 8, 1906 and continued weekly throughout the fall as long as the weather conditions permitted. The plants examined were : Common Red Clover [1] (Trifolium pratense), Mammoth Clover (Trifolium pratense perenne), White or Dutch Clover (Trifolium repens), Alsike Clover (Trifolium hybridum), Crimson Clover (Trifolium incarnatum), Alfalfa (Medicago sativa), White Sweet Clover (Melilotus alba), Hairy vetch (Vica hirsuta), Spring Vetch (Vica Sativa) and Soy Beans (Glycine hispida).

---

1. Scientific names as given by Britton.

During a thaw in the middle of January nearly all of the legumes previously mentioned were examined again and a number of plants of Black Medic (Medicago lupulina) were also examined. In the spring examinations will be made very frequently of all of the legumes mentioned.

In the green house work for the purpose of studying the life history of the nodules, I used the following legumes : Mammoth Clover, Alfalfa, Trifolium Alexandrian and Hairy Vetch. To determine the effect of a growing legume upon other plants of grass family growing adjacent, I used for the legumes Mammoth Clover and Soy Beans, and for the other plants not of the Leguminosae family I used Timothy (Phleum pratense) and Buckwheat (Fagopyrum Fagopyrum).

To my mind one of the most important objects of this study is to determine whether nodules die upon the roots of leguminous plants during the life of the host plant and are replaced by new ones, orwhether they continue to live until the death of the host plant. If the first condition is true then there can be but little doubt that the dropping off and decaying of the nodules will have a very decided beneficial action upon the other plants of the grass family growing adjacent. Whereas, if the latter condition is true, then a gramneous plant will receive little or no benefit from the legume until after it dies.

The study that I have undertaken is for the purpose of solving this very important question.

RESUME OF THE WORK BY OTHER INVESTIGATORS.

There is, of course, nothing new in the fact that after the growth of a leguminous crop, such as Soy Beans, Red Clover, and the like, that the soil is left in a better condition for the subsequent growth of a non-leguminous crop than it was before the legume was grown. In fact, the growth of such a leguminous crop is to a very great extent equivalent to the application of a nitrogenous manure for the cereal, or other non-leguminous crop. In deed, history tells us that more than two thousand years ago it was recognized by the Romans that the occasional growth of plants of the leguminous order had the effect of increasing the growth of the gramneous crops with which they were alternated; and in many instances it was stated that the effect was equivalent to that of manure.

The exact way by which this beneficial effect is brought about has long been a question of serious discussion. It is the purpose of the author to give a brief resume of the work that has been done along this line from ancient times down to the present.

From a historical point of view Schneider[1] has divided the subject as follows :

First Period.-- Initial study of leguminous root tubercles from Clos 1848 to Lawes and Gilbert 1860. During this period very little, if any, scientific investigations was done regarding the root tubercles of leguminous plants.

Second Period.--This period extended from Lawes and Gilbert investigations in 1860 to the investigations of Frank in 1879. During this period collateral investigations were made which led to the discovery of the true nature of root tubercles. The chief investigators of this period were Lawes and Gilbert of England and Hillriegel and Wilforth of Germany. The investigations made by these people pertained to the differences in the nitrogen supply and nitrogen assimilation of certain plants, as grasses, sugar beets, and leguminous plants. The final conclusion reached by Hillriegel was that there was a definite significant relationship between the root nodules and nitrogen assimilation of leguminous plants. Almost immediately these root tubercles were given marked attention which led to the discovery of their characteristic contents, namely, the bacteria.

Third Period.--This included investigations from Frank in 1879 to Schneider in 1893. During this period disputes and changes of opinion were not uncommon. It was not until the close of the period that anything like satisfactory

---

1.Minn. Bot. Studies 3 rd. Series, Part 2.

conclusions were reached regarding the true nature of the tubercles and the biological identity of the bacteria.

Fourth Period.--Thisperiod extends from the investigations of Schneider in 1893 up to about 1897. During this period preliminary investigations were made regarding the possible and practical utility of bacteria. The beginning of this period marks the first work done along agricultural lines. So far as is known the first paper outlining a course of research and giving a preliminary report of the work done with regard to the possible practical utility of bacteria in agriculture appeared in 1893.

Fifth Period.--This period overlaps into the previous one somewhat and includes the work that has been done along the economic value of bacteria and functionaly related organisms to modern agriculture.

Although there are many other classifications of this subject, this arrangement gives us a very good general idea of the development of this particular phase of the subject. It shows that our knowledge of this subject has passed through an evolution.

In 1866 the researches of Woroim[1] marked the beginning of serious investigations. Before this time the notions of early investigators regarding tubercles were probably empirical and derived more from superficial examination rather than from any serious attempt at an anatomical study of their structure and contents.

---

1. Mem.d l Acad. Imp. d'sciences VII 1866.

Chester[1] speaks of the works of Woroim and Erickson as follows : He (Woroim) found root nodules to be composed of a central portion of thin walled cells, of an outer ring and of an intermediate layer or vascular ring. In the central portion the contents were cloudy and closer examination showed the cells in this portion to be filled with peculiarly shaped bodies, which were sometimes rod-like, or at other times, forked and presenting a variety of forms. Woroim regarded them as causative agents in the formation of the tubercles, although he failed to offer any proof of this assumption.

In 1874, Erickson found in the newly developed tubercles long, branching filaments resembling the mycelium of fungus. these threads he considered to be the infecting agents. Later on in the development of the tubercles, he observed the presence of the bacteroides, noted by Woroim, but failed to connect the two as corelated structures.

Thus, it will be seen that in the early observations of Woroim and Erickson, we have the germ and substance of all that has been discovered since regarding root tubercles. Both recognized as is held by the majority of investigators of to-day, the two classes of bodies, the filamentous and the bacteroid.

In 1889, Atwater and Woods[2] reported results from experiments with alfalfa,peas, and with cereals that had been

---

1.Penn. Dept. Agr. Bul. 98.

2. Jour. Chem. Soc. 1891; also Amer. Chem. Jour. 12.

conducted for several years. These experiments point to the following conclusions :

1. That atmospheric nitrogen is undoubtedly acquired during the growth of peas and alfalfa, and that the amount of nitrogen gained increases with the number of root tubercles.

2. That cereals do not manifest the power of acquiring nitrogen from the air nor are tubercles formed on them, as in the case of the leguminous plants.

3. That as a rule, the greater the abundance of root tubercles in these experiments the larger and more vigorous were the plants and the greater was the gain of nitrogen from the air.

4. A loss of nitrogen occured where there was no tubercles formed.

In 1889, Lawes and Gilbert were able to state from experiments made by them that separate analyses of nodules of different character or in different conditions, showed that while some were more or less exhausted and contatined a low percentage of nitrogen, others contained a higher percentage. It was thought that the latter were new and active. Thus, the results pointed to the pointed to the interesting conclusion that, although with the plant of longer life, the sunflower, the earlier formed nodules became exhausted, others were constantly produced to provide for future growth.

Albert Schneider[1] has made several observations upon

---

1. Bot. Gazette Vol. 36, No. I.

the winter and early spring condition of root tubercles. He arrives at the following conclusions :

1. A considerable number of bacteria of biennial and perennial plants forming root tubercles are killed during the winter months.

2. Root tubercles of perennial herbaceous legumes attain their full growth during the early part of the first season.

3. Most root tubercles of perennial herbaceous legumes die or decay at the close of the second season, returning only a part of the bacteria to the soil.

The bacteria that are distributed in the soil by the decaying of the plant or of the tubercles gain admission to the new host plant through the root hairs and require but a short time to increase greatly in number. The irritation thus caused in the tissues of the young rootlet causes it to take on an altered form at this point. That is, a nodule is formed. These nodules vary from minute spheres to swellings as large as marbles, while the lobed or compound nodules often occur in clusters much larger.

Although the size and external appearance of nodules vary greatly, it can hardly be stated that each legume has a characteristic shaped nodule that distinguishes it from other legumes. In the case of peas, they occur generally as conglomerations; on the roots of vetches the nodules are generally single. Lupines appear to have two kinds of tubercular development, the most prevalent being swellings which entirely encase the thick roots. Alfalfa appears to have three forms of nodules.

One form appears as warty excressences mostly near the neck; another form as single nodules on small roots; and still another form as united into large colonies. The first form is found at shallow depths. The second form is found at three to five feet below the surface, and the third type is found at all depths. Further observation has led to the conclusion that the size, number and distribution of the nodules depends very greatly upon the kind of soil in which the legume grows.

A series of experiments was commenced at Rothamsted[1] in 1890 in which four annuals, peas,beans, vetches and yellow lupines and four plants of longer life, white clover, red clover, sainfoin, and alfalfa, were grown in specially made pots, so arranged that some of the plants of each description could be taken up and their roots and nodules studied at successive periods of growth. The annuals were examined three times and the plants of longer growth four times. The annuals were examined first, when active vegetation was well established, second, when it was supposed that the point of maximum accumulation had been reached, and third, when nearly ripe. The plants of longer life were examined at the end of the first year, and in the second year, when active vegetation was reestablished, when the point of maximum accumulation had been reached, and lastly, when the seed was nearly ripe.

Each of the eight kinds of plants were grown in sand (with the plant ash) watered with the extract from a rich soil; also in a mixture of two parts rich garden soil and one part sand.

---

1. Rothamsted Report 1895.

These plants were grown in the air, but were protected somewhat from the heavy rain. The observations taken at the different times stated show that in the sand the infection was comparatively local and limited, but some of the nodules developed to a great size on the roots of the weak plants so grown. In the rich soil the infection was much more general over the whole area of the roots, the nodules were much more numerous, but generally very much smaller. Eventually the nodules were picked off the roots, counted, weighed and the dry substance together with the nitrogen in them determined.

The table on the following page shows the results obtained from these observations. The plants given in the table showed the most normal development of any grown.

	Date of taking up.	No. of plants grown.	N O D U L E S Approx. No.	Wt. dried at 100°C. grams.	N I T R O G E N In dry. %	Actual. grams
Peas						
In Sand:	1890					
1st Per.	Aug.4.	3	253	0.229	6.630	0.0152.
2nd "	Sept.24.	3	335	.516	3.593	.0185.
3rd "	Nov.29.	3	328	.162	2.164	.0034.
In Soil:						
1st Per.	Aug.5.	3	324	.743	5.022	.0373.
2nd "	Sept.26.	3	1,353	1.497	3.167	.0474.
3rd "	Dec.3.	3	1,513	1.600	2.797	.0447.
Sainfoin						
In Sand:	1890-91.					
1st Per.	Dec.10.	3	82	.153	7.346	.0112.
2nd "	May 15.	3	148	.229	5.793	.0133.
3rd "	Je.12.	3	360	1.043	6.151	.0641.
4th "	Sept.11.	3	2,891	4.403	4.735	.2085.
In Soil:						
1st Per.	Dec.13.	3	226	.040	6.259	.0025.
2nd "	May 15.	3	2,018	1.492	6.286	.0937.
3rd "	Je.12.	2	2,125	.649	6.363	.0412.
4th "	Sept.14.	3	3,413	3.299	7.066	.2331.

It is seen from the above table, that, at the third period of growth of the peas in sand, the amount of dry matter of the nodules was very much diminished, the percentage of

of nitrogen in the dry matter was very much reduced and the actual quantity of nitrogen remaining in the total nodules was also very much reduced. In fact the nitrogen of the nodules was almost exhausted. The peas grown in rich soil, however, maintained much more vegetative activity at the conclusion and showed a very great increase in the number of nodules from the first to the third period. With this there was also much more dry substance and even a greater actual quantity of nitrogen in the total nodules at the conclusion. Still, as in the peas grown in sand, the percentage of nitrogen in the dry substance of the nodules was very much reduced at the conclusion.

In the case of the plant of longer life, the sainfoin, there was, both in sand and in the soil, very great increase in the number of nodules and in the actual amount of dry substance and of nitrogen in them as the growth progressed. The percentage of nitrogen in the dry substance of the nodules also showed, even in the sand, comparatively little reduction, and in the soil even an increase. In fact, separate analyses of nodules of different character or in different conditions showed that while some were more or less exhausted and contained a less percentage of nitrogen, others contained a high percentage and were doubtless new and active. Thus the results pointed to the interesting conclusions that in the case of the annual when seed is formed and the plant more or less exhausted, both the actual amount of nitrogen in the nodules and its percentage in their dry substance are

greatly reduced, but that with the plant of longer life, although the earlier formed nodules became exhausted, others are constantly produced, thus providing for future growth. The results further show that there is intimate connection between the gain of nitrogen by Leguminosae and the development of nodules on their roots.

The plants grown in sand had much fewer nodules than those grown in the rich soil. The size of the nodules, however, was much greater on the roots of the plants grown in the sand.

The smaller number of nodules found on the plants grown in sand may be due to the fact that the infection was dependent on the additions of rich soil extract. The diffusion of the bacteria might be only limited, and the infection of the roots only local and accidental. The much greater size of the individual nodules may be due to the want of power in the more weakly plant growing in nitrogen-free soil to resist the free development of the parasite. On the other hand, in the mixture of rich soil and sand the bacteria would probably be distributed throughout it and the roots accordingly exposed to infection along their whole range.

In nearly all the experiments that have been conducted up to the present time, more nodules have been found to be developed on the roots of plants that were grown in soils deficient in nitrogen than on those grown in soils rich in nitrogen. Observations made at the West Virgina Agr. Exp. Station[1], however, do not agree with the results obtained

---

1. West Virgina Exp. Sta. Bul. 105.

at most other places. In examining a number of alfalfa plants on the various experimental plats, one thing that was observed in particular was the abundance of small tubercles on a few large plants with their roots in clods of well rotted manure or of sod, thus indicating that material of this kind was not only favorable for the growth of alfalfa, but also for the formation of tubercles.

These observations correspond exactly to the observations made by the author upon the experimental plats on the University farm. Wherever a piece of partially decayed humus was found, there was almost invariably an abundance of nodules present, and usually in the form of clusters.

LIFE HISTORY OF NODULES AS FOUND IN THE FIELD.

The field work to determine the life history of nodules upon legumes was commenced Oct. 8, 1906 and continued throughout the fall until Nov. 12, 1906. Durind this period of six weeks, weekly examinations were made of the following legumes; Common Red Clover, Mammoth Clover, White Clover, Alsike Clover, Crimson Clover, Alfalfa, White Sweet Clover, Hairy Vetch, Spring Vetch and Soy Beans. These plants were all found growing in greater or less abundance upon the University farm.

In an examination of the nodules upon the roots of the different legumes no attempt was made to give the actual number of nodules upon any given plant. On account of the great difficulties involved in removing the entire root system of the plant from the soil, it was thought best to give a comparative statement of the number of the nodules

rather than exact numbers. In removing the plants from the soil every precaution was taken to remove the entire root system. A deep narrow hole was made by the side of each p plant removed. Then with the spade on the other side of the plant the soil and and plant were pushed into the excavation made. By following this method I found that I was able to remove a plant from the soil with a large proportion of its roots intact. With the exception of the old alfalfa plants a large proportion of the roots were removed from the soil and carefully examined in each case. In examining the nodules upon the roots of the old alfalfa plants, I found it impracticable to attempt to remove the entire root system. Consequently with the alfalfa plants the results obtained apply only to the upper three feet of the root system.

The number of plants examined in each case depended somewhat upon the size of the plant and the difficulty involved in removing it from the soil. In the case of the smaller plants, ten of each were removed from the soil at one time and an average of these ten plants was taken. With the larger deeper rooted plants, like alfalfa for example, only two or three of the plants were removed for examination.

Of the plants examined all were in a green thrifty, apparently growing condition during the whole period of observation, with the one exception of the soy beans. These died or ripened long before the completion of the field work.

The following tables show the age of the plants examined, number, size and condition of the nodules on each legume at the different times examined.

Oct. 8, 1906.

Name	Age	Number	Size	Condition
Red clover	Over 1 yr.	Few	Med.	Old
Red "	Under "	Many	Small	Young
Mammoth "	Over "	"	"	Med.
White "	" "	"	"	Young
Alsike "	" "	"	Med.	"
Crimson "	" "	Few	"	Old
White Sweet	" "	Very many	"	Old and young
Alfalfa	Over 3 yr.	Very few	Clusters	" " "
Alfalfa	Over 1 yr.	Few	"	" " "
Alfalfa	Under 1 yr.	Many	Small	Young
Hairy Vetch	Over 1 yr.	"	Large	Old
Hairy "	Under 1 yr.	"	"	Young
Spring "	Over 1 yr.	Few	Med.	Old
Spring "	Under 1 yr.	Med.	"	Old & young
Soy beans	" "	Ver many	Very large	All old

Oct. 15, 1906.

Name	Age	Number	Size	Condition
Red clover	Over 1 yr.	Med.	Small	Healthy
Red "	Under "	Many	"	"
Mammoth "	Over "	Med.	"	Old
White "	" "	Many	"	Healthy
Alsike "	" "	"	Med	"
Crimson "	" "	Few	Small	Old
White Sweet	" "	Many	Med	Mostly old
Alfalfa	Over 3 yr.	Very few	Clusters	Old & Young
Alfalfa	Over 1 yr.	Few	"	" "
Alfalfa	Under "	Many	Small	Young
Hairy Vetch	Over "	"	Med.	Old
Hairy "	Under "	"	Large	Young
Spring "	Over "	Few	Med.	Old
Spring "	Under "	"	"	Old & Young
Soy beans	Under "	Very many	Large	Very Old

Oct. 22, 1906.

Name	Age	Number	Size	Condition
Red clover	Over 1 yr.	Med.	Small	Old
Red "	Under "	Many	"	Healthy
Mammoth "	Over "	Few	Med.	Old
White "	" "	Many	Small	Healthy
Alsike "	" "	"	Med.	"
Crimson "	" "	Few	Small	Old
White Sweet	" "	Many	Med.	"
Alfalfa	Over 3 yr.	Very few	Clusters	"
Alfalfa	Over 1 yr.	Few	"	"
Alfalfa	Under "	Many	Med.	Young
Hairy Vetch	Over "	"	"	Old
Hairy "	Under "	"	Large	Young
Spring "	Over "	Few	Med.	Old & Young.
Spring "	Under "	Few	"	" "
Soy beans	Under "	Many	Large	Badly decayed

Oct. 29, 1906.

Name	Age	Number	Size	Condition
Red clover	Over 1 yr.	Few	Small	Old
Red "	Under "	Many	"	Healthy
Mammoth "	Over "	Few	"	Old
White "	" "	Many	Small	Old & Young.
Alsike "	" "	Med.	Med.	" "
Crimson "	" "	Few	Small	Old
White Sweet	" "	Many	Med	Old
Alfalfa	Over 3 yr.	Very few	Clusters	"
Alfalfa	Over 1 yr.	Few	"	"
Alfalfa	Under 1 yr.	Many	Med.	Young
Hairy Vetch	Over 1 yr.	"	Large	Old
Hairy "	Under 1 yr.	"	"	Young
Spring "	Over 1 yr.	Few	Med.	Old
Spring "	Under 1 yr.	"	"	Old & Young
Soy beans	Under 1 yr.	Many	Large	Badly decayed.

Nov. 5, 1906.

Name	Age	Number	Size	Condition
Red clover	Over 1 yr.	Few	Small	Old
Red clover	Under "	Many	"	Healthy
Mammoth "	Over "	Few	"	Old
White "	" "	Many	"	Old & young
Alsike "	" "	Med.	"	" "
Crimson "	" "	Few	"	Old
White Sweet	" "	Many	Med.	"
Alfalfa	Over 3 yr.	Very few	Clusters	"
Alfalfa	Over 1yr.	Few	"	"
Alfalfa	Under 1 yr.	Many	Med.	Healthy
Hairy Vetch	Over 1 yr.	"	Large	Old
Hairy "	Under "	"	"	Old & young
Spring "	Over "	Few	Med.	Old
Spring "	Under "	"	"	Mostly old
Soy beans	Under "	Mostly gone		

Nov. 12, 1906.

Name	Size	Number	Size	Condition
Red clover	Over 1 yr.	Few	Small	Old
Red "	Under 1 yr.	Many	"	Mostly old
Mammoth "	Over 1 yr.	Few	"	" "
White "	" "	Many	"	" "
Alsike "	" "	Few	"	" "
Crimson "	" "	"	"	" "
White Sweet	" "	Many	Med.	" "
Alfalfa	Over 3 yr.	Very few	Clusters	Very old
Alfalfa	Over 1 yr.	Few	Small	Old
Alfalfa	Under 1 yr.	"	"	Healthy
Hairy Vetch	Over 1 yr.	Many	Large	"
Hairy "	Under 1yr.	"	"	"
Spring "	Over "	Very few	Med.	Old
Spring "	Under "	Few	"	"
Soy beans	" "	Entirely gone		

It will be seen from the preceeding tables that in general the nodules remained about the same throughout the period of observation. In some instances, as for example, with the alfalfa nodules, the nodules decreased in number toward the end of the period. On Nov. 12, it was almost impossible to find any nodules on the roots of the old alfalfa plants. The roots of the young alfalfa plants were well covered with heaalthy nodules throughout the six weeks. From the tables, it will also be noticed, that there were fewer young nodules on the roots of the plants toward the end of the period of observation, than at the beginning. With the soy beans no young nodules could be found even at the beginning. As soon as the plants were frosted and began to die the nodules also began to show signs of decay. About the third week after the plants were injured by the frost the nodules were badly decayed. The fifth week most of the contents of the nodules were badly decayed and their contents was of a whitish slimy fluid. At this time very few of the nodules could be found entire. In most cases only the outside thin cases were left. These looked perfectly dead.

Contrary to the opinion reached by most investigators, that more nodules are borne upon legumes grown in soils depleted in nitrogen, the writer was able to find an abundance of nodules on plants grown in soils rich in humus. In many cases, in soils containing little humus it was almost impossible to find any nodules at all. This was particularly

true with alfalfa plants. On uninoculated plats on the University Experimental grounds, that were used as check plats to determine the effect of different kinds of innoculation upon nodule formation, the writer made several observations of the roots of the growing alfalfa. In no case was he able to find nodules on the roots of these plants unless he foun a small piece of partially decayed humus. On the other hand, whenever he found a bit of partially decayed humus either rotted sod or manure, he was able to find a large cluster of compound nodules. Many times the clusters of nodules were entirely within the humus and could not be seen at all until it was broken apart.

On January 14, 1907, field observations were made of the following legumes: Common Red Clover, White Clover, Alfalfa, Hairy Vetch, Spring Vetch and Black Medic. In every case the roots quite well covered with nodules. The most of the nodules on all of the plants, except the Black Medic, were brown and looked as though they were in a dormant condition. In each case, however, some young nodules were found on the very small rootlets. The roots bearing these young nodules looked as though they themselves were young.

The roots of the Black Medic examined at this time were well covered with young nodules. These were also borne on the young rootlets.

It may be stated that at this time the ground was nearly free from frost. No difficulty was experienced in removing

the plants from the soil on account of frost. All the flants had some green leaves. They looked as though they had just started to grow. The Black Medic showed the most new growth of any of the plants examined.

Observations made at frequent intervals after January 14th seemed to verify the same facts. Just as soon as new growth was produced nodules were formed upon the new rootlets.

GREEN HOUSE STUDY OF NODULES

The green house work was commenced on Nov. 19, 1906 and continued until the last of April.1907. The legumes used for the purpose of studying the life history of their nodules were: Alfalfa, Vetch, Mammoth Clover and Alexandrian clover.

Three methods have been persued to study the nodules upon these plants. (1) The seeds were sown in ordinary boxes and the plants removed from time to time to determine the number, size and condition of the nodules. (2) Boxes of glass sides were used and observations made without removing the plants from the soil. (3) Plants were grown in water cultures.

In the first method the seeds were sown in a rather light very fine sandy soil. After the plants had been up about one week a few were removed from the soil and examined every few days. This was done so carefully that the plants did not show any effects of their removal from the soil. By fol-

lowing this method the writer was able to tell when the first nodules appeared and the rapidity with which they increased after they did appear.

The drawings at the end of this work as well as the following table show the results of these observations.

Name	Time when sown	Appearance of first nodules	Number on plant
Alfalfa	Dec. 11, '06	Jan.7,'07	2 on 10 plants
Vetch	Dec. 7,'06	Jan.3,'07	1 to 3 "
Alexandrian clover	Jan. 7,'07	Jan. 21,'07	1 to each "
Mammoth clover	Nov. 23, '06	Dec. 17,'06	1 to " "

It will be noticed that, with the exception of Alexandrian clover, it required nearly one month after sowing before any nodules were developed. Even at the end of one month comparatively few nodules were developed. Perhaps this can partially be explained by the slow growth made by the plants. During the first month and a half of the experiment the temperature in the green house was never above 60° F and it frequently fell to below 40°F. This low temperature naturally retarded the growth of the plants very much. After the temperature was raised to above 70°F the plants grew much faster and developed more nodules.

The following table shows the increase of nodules due to the age and growth of the legumes.

	Jan.28	Feb.4	Feb11	Feb.18	Feb.25	Mar.4	Mar.20
Alfalfa	3	5	6	10	11	13	16
Vetch	8	13	15	15	17	17	18
Alex. clover	1	3	8	17	18	20	22
Mammoth "	8	20	32	35	37	40	42

From the above table it will be seen that the percentage increase was much greater during the first few weeks directly after the first appearance of nodules, than after the plants had reached greater size. The formation of new nodules appeared to be almost entirely on the new roots, scarcely any appearing on the old roots. When the nodules first appeared on the young rootlets it was sometimes very difficult to distinguish between them and young rootlets. In some cases swellings that appeared to be nodules at first later developed into roots.

On Feburary 18, 1907 some of the older nodules borne near the surface of the soil on the main roots were noticed to be decreasing in size. These have been closely watched since and many have been found to have entirely disappeared. Others are very small and are noticed now only by a slight bulging. The writer's observations of the plants grown in boxes with glass sides seem to verify this same result.

Thus, instead of decaying and falling off from the host

plant the writer is of the opinion that they are used, at laest those on the main roots, by the host plant.

The second method used to determine the life history of the nodules, namely; theoone in which the plants were grown in boxes with glass sides, was not as satisfactory as had been wished. Although black paper was used to obstruct the light, the roots did not seem to go toward the side light to any great extent. In several cases. however, the roots did go to the surface of the glass and nodules were formed where they could easily be viewed. No attempt was made to count the number of nodules upon any particular plant. This was impossible because not enough of them were in sight to be of any use in this respect. Individual nodules, however, were viewed every few days. It was observed, that with the simple nodules, they reached their full development within a very few days after they were formed. In most cases it only took three or four days for them to reach their full size. On the other hand, with the compound nodules, that is, those that were lobed, a much longer time was required. In several cases the writer has observed that some of the nodules on the main roots have been found to be reabsorbed by the host plant, and in no case has he observed a single one to fall away from the host plant. Thus, this observation corresponds exactly with the conclusions reached where the plants were grown in ordinary boxes and removed frequently from the soil.

The third method used to determine the life history of nodules upon legumes was to grow the plant in a water culture. Instead of growing the plants from the seed in the solution young plants with nodules already upon them were taken for the experiment. Two plants of Vetch, Mammoth clover and Alfalfa were used.

The soil solution used was made by taking 500 cc. of water and 100 grams of soil. They were placed together and stirred for three minutes, then allowed to stand until wanted for use.

The plants were placed in bottles about 8 inches high and 2 and 1/2 inches in diameter. These bottles were covered with black paper to keep out the light. The plants were supported in the bottles by notches in the sides of cork. After the corks were fitted to the bottles and the notches in the side made to admit the plants, they were dipped in warm paraffin. The plants were held upright in the bottles by packing cotton around them where they went through the corks. One plant was placed in each bottle. The soil solution was changed twice a week.

The following table gives the results of the experiment. It might be well to state that the plants in this solution did not grow nearly as well as those in the soil. The Mammoth Red clover made the most normal growth and the Vetch the poorest.

	Number of nodules at first, Feb. 9.	Feb. 16	Feb. 25	Mar. 4.	Apr 10
Alfalfa I	2	4	4	4	6
Alfalfa II	2	4	5	5	5
Mammoth clover I	15	22	22	43	89
Mammoth clover II	20	23	29	45	110
Vetch I	8	10	12	12	20
Vetch II	10	14	17	24	30

From the above table it will be seen, that with the exception of Alfalfa, the number of nodules is steadily increasing. This increase is very largely on the young roots. Some of the oldest nodules along the main root have shriveled up and are apparently being absorbed by the host plant. None of the nodules on the roots of the plants grown in water culture have fallen off from the plant.

EFFECT OF A LEGUME UPON OTHER PLANTS GROWING ADJACENT.

Within the last few years there has been a growing tendency among the farmers, that certain legumes, when grown in connection with other plants not of the leguminose family, produce beneficial effects upon the non-leguminose plants. As an illustration of this fact they usually cite the good growth of corn when grown with Soy Beans and the fine growth of oats when grown with peas. Many farmers of the central part of New York State have so much faith in the beneficial effect from the legume that they seldom plant a field of corn without planting Soy Beans with it. This is particularly true when the corn is intended for ensilage purposes. The same thing is true with oats when they are wanted for soiling or for hay. Peas are nearly always sown with the oats with the idea that the oats will make a much larger growth than they would were the peas not present.

It is also noticed that Kentucky Blue grass grows much

better under Locust trees than under any other trees, thus leading one to believe that the extra growth of the grass under the locust trees is due the leguminous effect of the locust.

At the Ontario Agricultural College and Experiment Farms[1], it was found that a larger yield of oats and peas were produced when sown together, than when the oats were sown alone.

The beneficial effect of legumes is even more emphasized in the older countries that have been cropped for many centuries. In India, for example, the natives seldom attempt to raise a crop of corn without planting alternate rows with the legume, Cajanus indicus. The benefit of this legume upon the growth of corn has become so well known that it is the universal practice to devote every other row to it throughout the entire corn field.

These observations of the farmers have attracted the attention of investigators and experimenters. Investigators are anxious to prove or disprove the very common idea that legumes are beneficial to non-leguminous crops.

In the spring of 1906 a field of corn was planted on the University Farm with the idea of determining the effect of a legume, Soy Beans, upon the growth and yield of the corn. The soy beans were planted in every alternate hill of corn. By this means an excellant opportunity was afforded to compare the corn grown alone and that grown with soy beans in the hill. It was thought, that by this method of planting, any difference in the growth of the corn would be due to the

soy beans entirely and not to other conditions, such as soil, cultivation and etc.

In the fall just before the crop was harvested, careful observations were made by the writer to see if the soy beans had any effect upon the growth of corn. Although the soy beans had made a very good growth, and the roots were almost completely covered with large nodules, no direct benefit could be seen in the growth of the corn in the hills supplied with the legume. On the other hand, the corn did not show that the soy beans were detrimental. In both cases the corn made an excellant growth. It is possible that different results would have been obtained, had the experiment been c carried on, on soilmore deficient in nitrogen. The soil used for the test is considered quite productive. Nearly every year it produces good crops. This particular experiment, however, failed to show the beneficial effect that is generally attributed to a growing legume.

GREEN HOUSE WORK TO DETERMINE EFFECT OF A LEGUME

For this work Mammoth clover was used with Timothy and Soy Beans with Buckwheat. To determine the effect of the clover upon the growth of the timothy, timothy and clover were sown together and each was sown separately. A duplicate in each case was also made. That is, in all, six boxes were used, two containing timothy alone, two containing clover alone and two containing both timothy and clover. To determine the effect of the soy beans upon the buckwheat the buckwheat and soy beans were sown together in two boxes; then in two other boxes with partitions in the middle, each was sown separately; in two boxes similar to the last two, but without partitions, each was sown separately exactly the same as in the boxes with partitions.

Although these experiments have not been conducted long enough to draw definite conclusions, yet in the case of the

timothy, it is noticeably better where grown with the clover. The individual plants look more thrifty. They are larger. The leaves are wider and of a darker green when grown with the clover than when grown alone. Fewer dead leaves are found on the timothy plants grown with the clover than on those grown alone.

Since the soil, moisture and heat were as nearly identical in both cases as it was possible to have them, it does not seem improbable that the increased vigor which the timothy plants in the clover appear to have is due entirely to the beneficial effects of the clover.

In the case of the influence of the soy beans upon the growth of the buckwheat, the results thus far do not prove conclusively that the buckwheat has received benefit from the soy beans. Further development of the plants will be necessary before definite conclusions can be drawn.

CONCLUSIONS

From the observations of the writer and the experiments conducted by other investigators, the following conclusions seem warranted:

1. Under ordinary conditions new nodules are produced on the rootlets of leguminous plants during the entire active growing period of the plant.

2. Unless the plant is partially destroyed by some external factor, as mutilation, frost, etc., few, if any, of the nodules drop off from the roots until the death of the host plant.

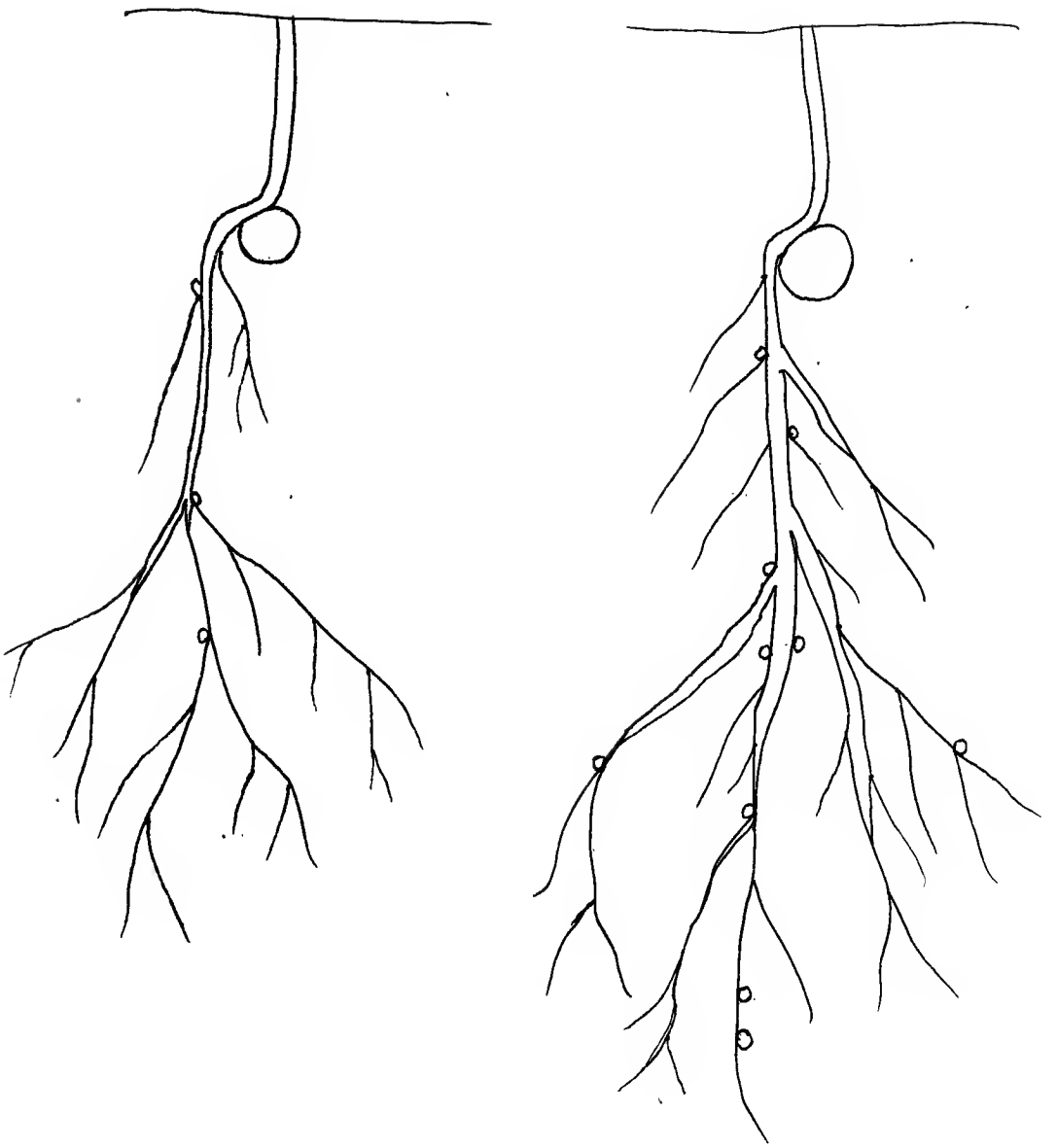
3. The majority of the nodules formed remain on the roots of the host plant until the latter dies. Some, however especially along the main roots, are apparently reabsorbed. Moore [1] found that the high percentage of nitrogen in nodules was just before flowering and the formation of fruit. He also found that the contents of most of the nodules disappeared as the plant reached maturity and the inclosing tissue shriveled up.

---

1. Bulletin No. 71 Bureau of Plant Industry.

4. At the present time there is insufficient data at hand to form definite conclusions in regard to the effect of a growing legume upon other plants of grass family growing adjacent. In the experiment carried on by the writer with red clover and timothy, the timothy showed a decided benefit in favor of the clover growing adjacent. On the other hand, experiments conducted on the University farm with corn grown in connection with soy beans (See pages 33 & 34) did not show any beneficial effect of the legume.

Thus, it will be seen, that more data on this subject will be required before satisfactory conclusions can be drawn.



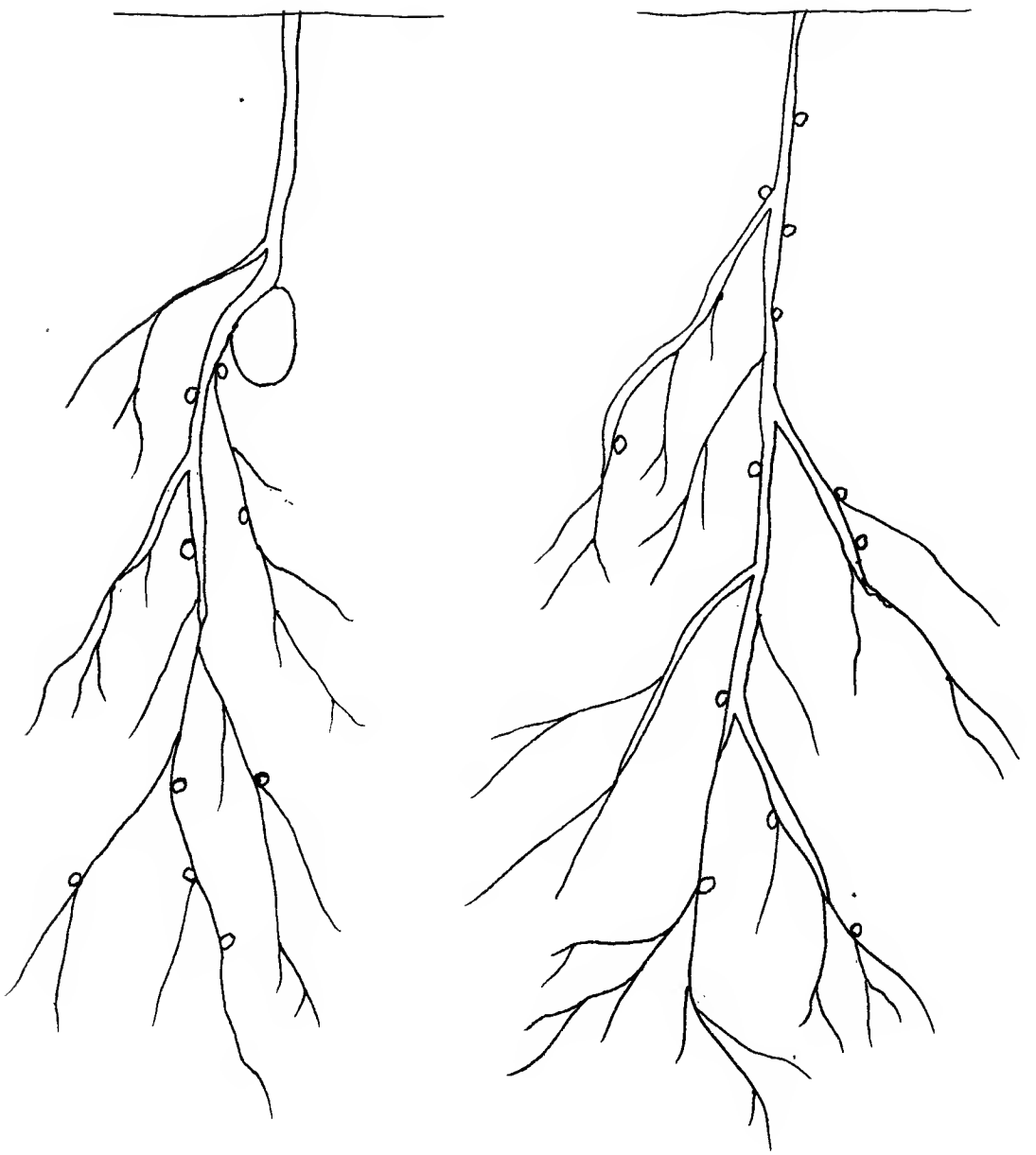
Vetch (Vicia sativa)

Enlarged twice.

27 days after planting.

Enlarged twice.

38 days after planting.



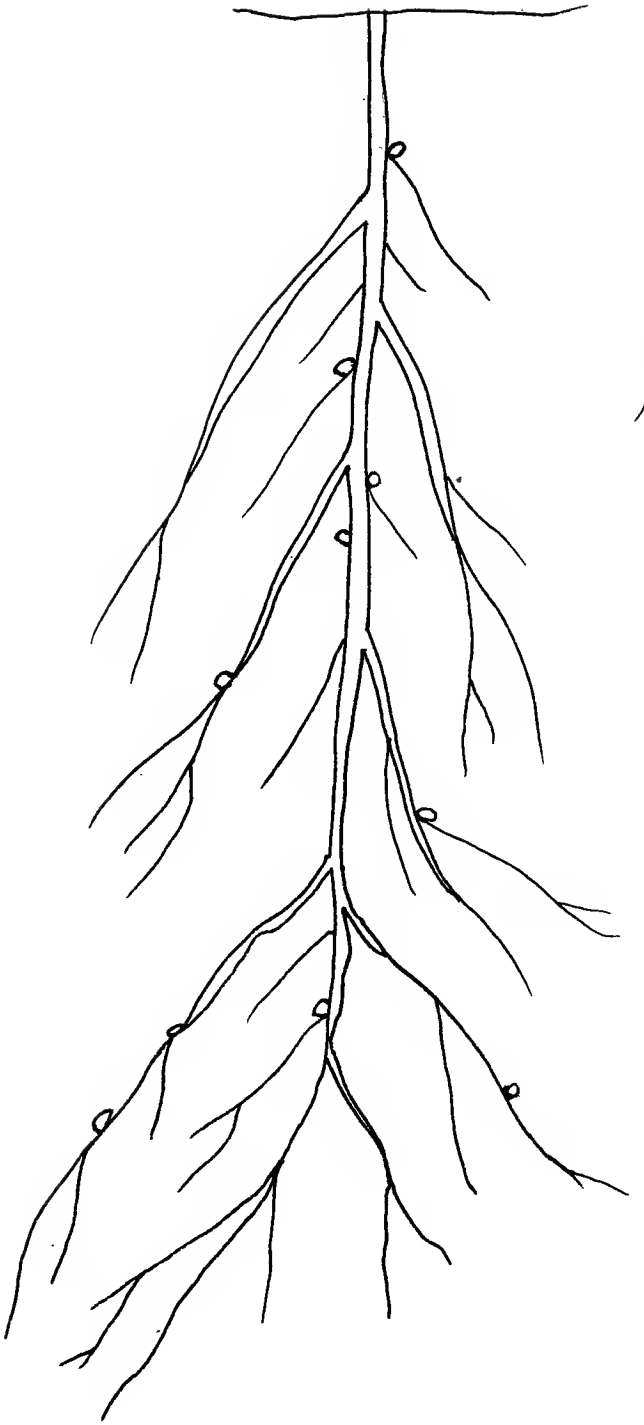
Vetch (Vicia sativa)

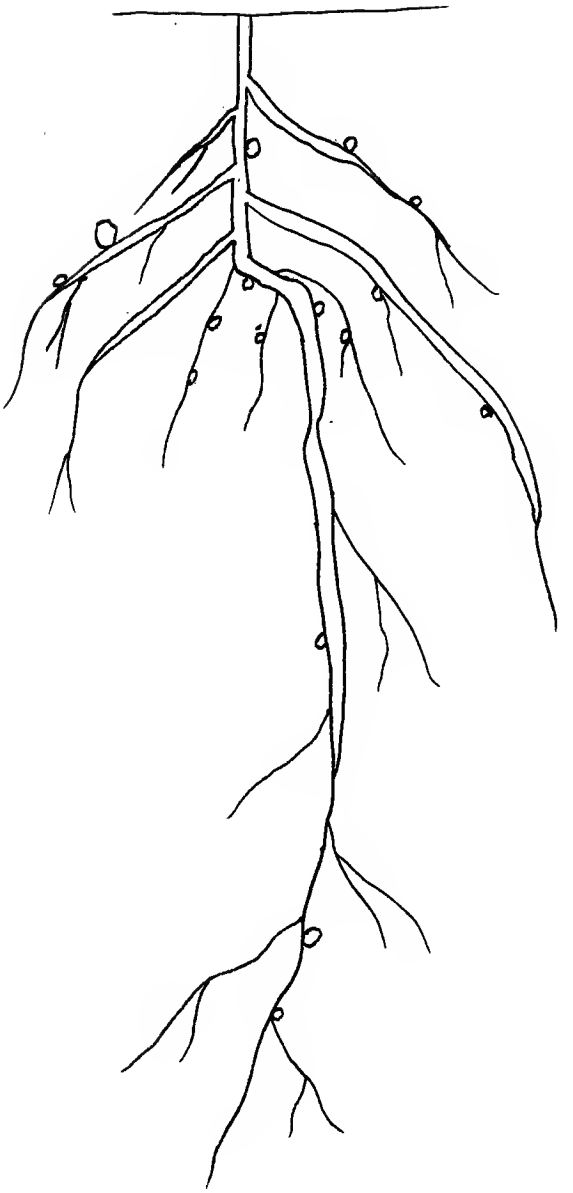
Enlarged twice.

43 days after planting.

Enlarged twice.

60 days after planting.

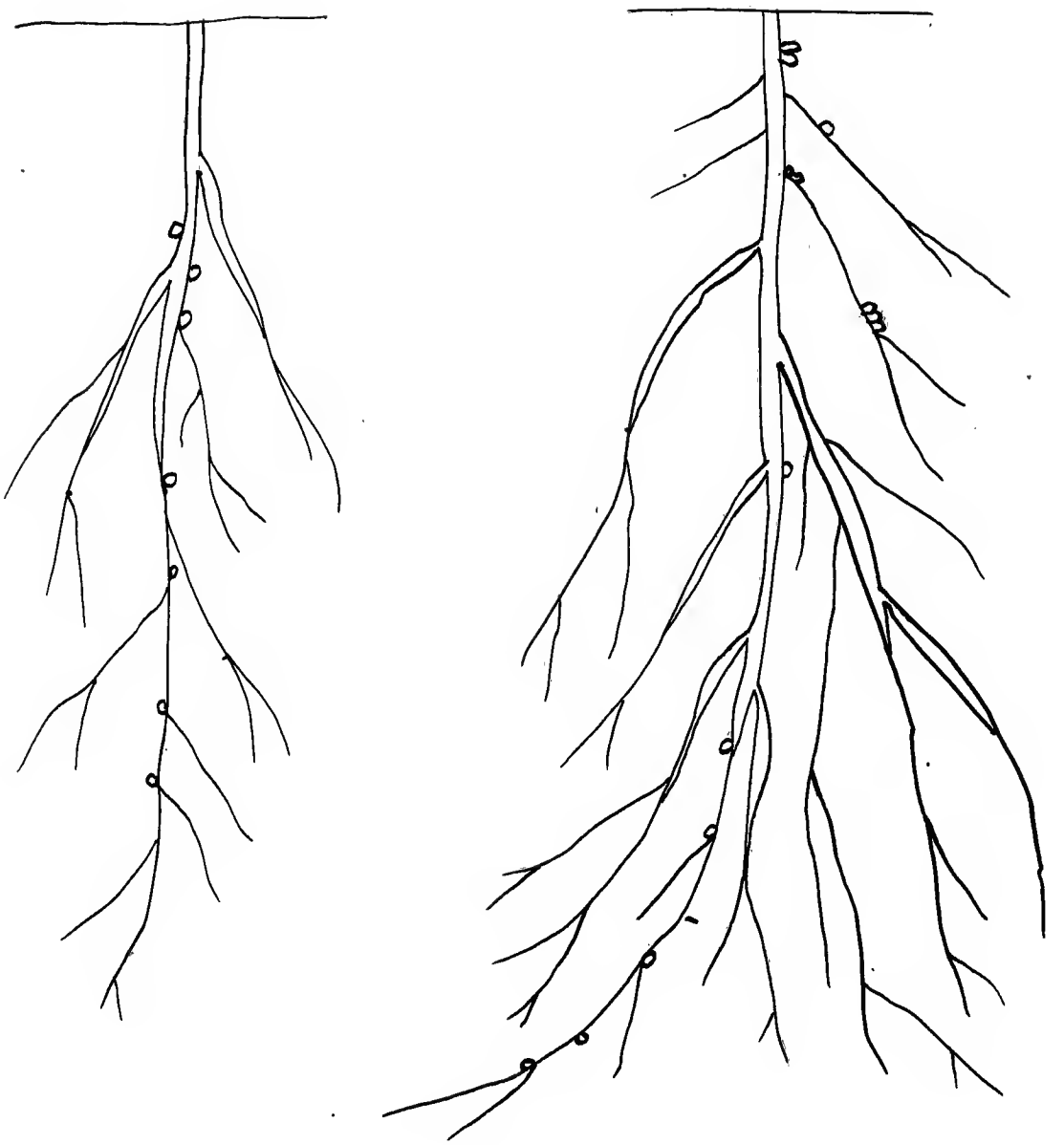




Vetch (Vicia sativa)

Enlarged twice.
75 days after
planting.

Enlarged twice.
87 days after
planting.



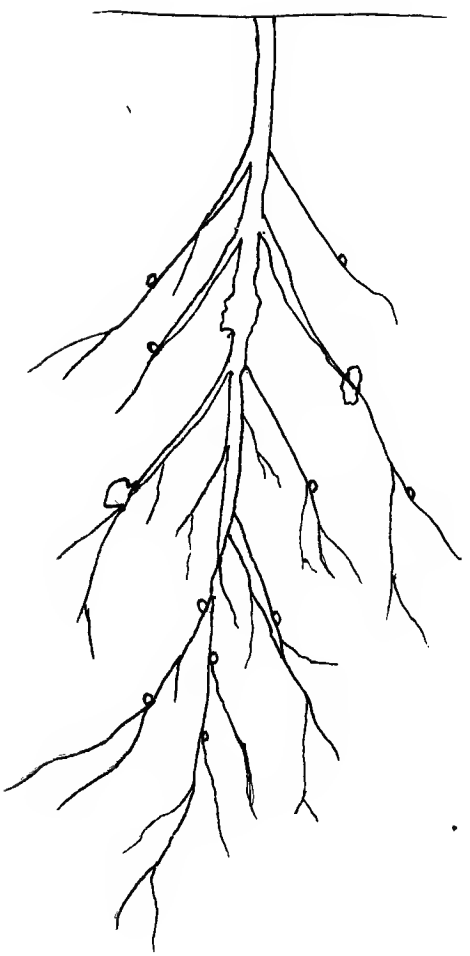
Alexandrian clover.

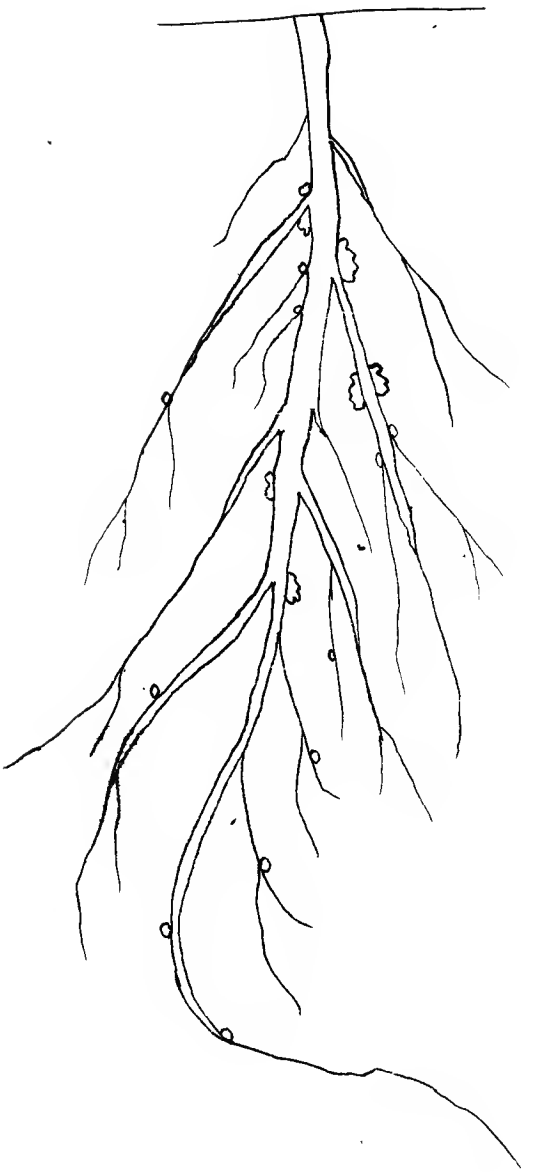
Natural size.

44 days after planting.

Natural size.

52 days after planting.





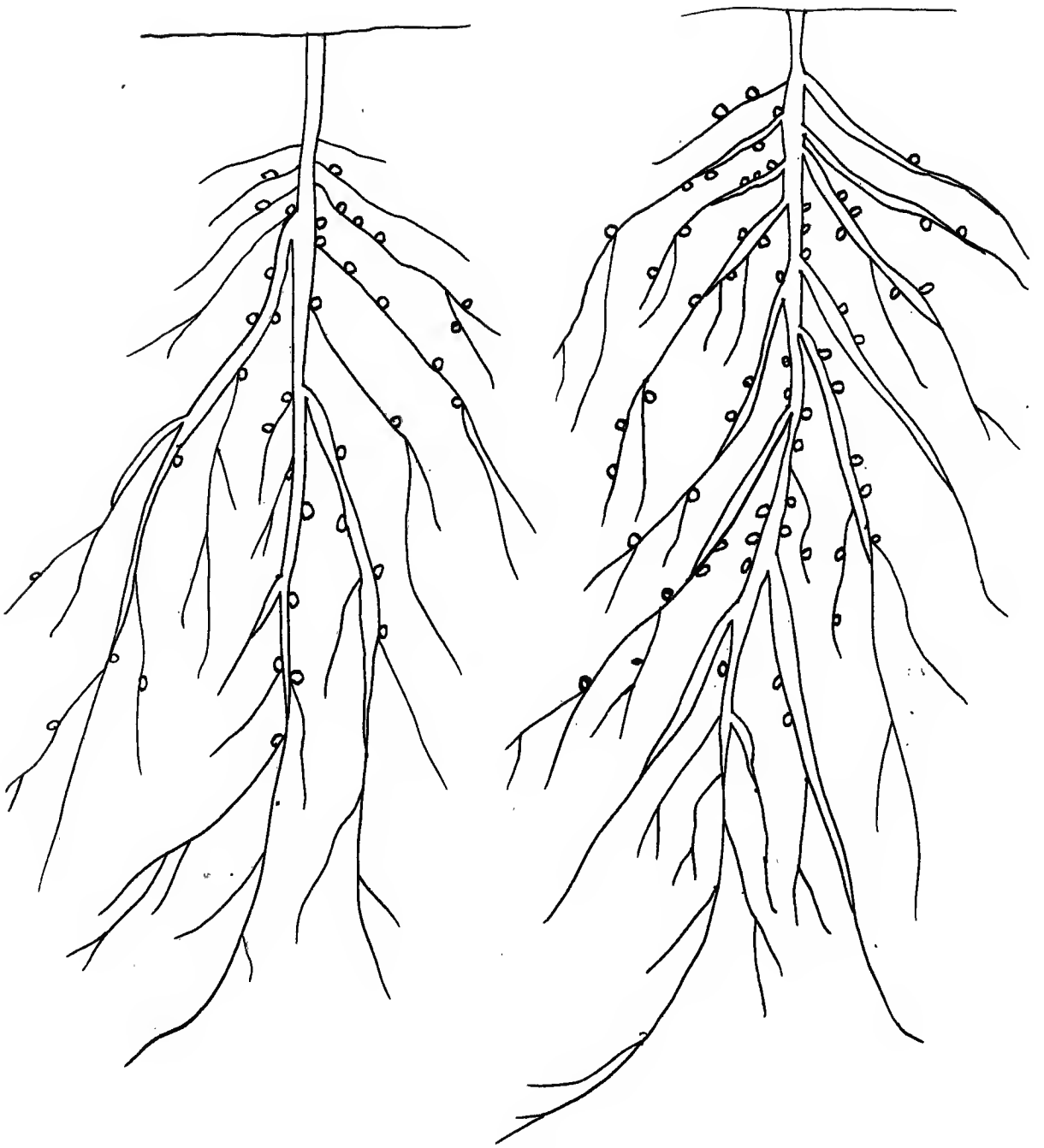
Alexandrian clover.

Natural size.

56 days after planting.

Natural size.

94 days atfer planting.



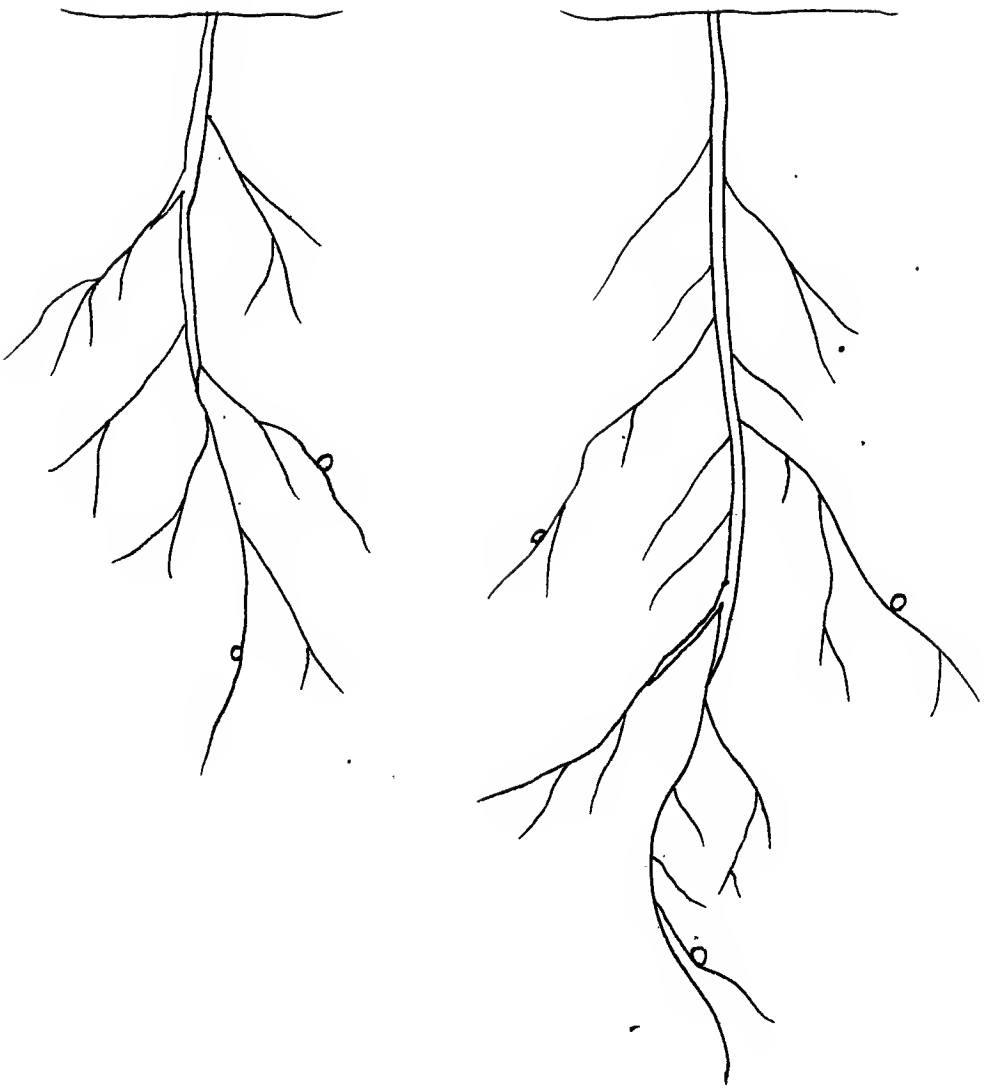
Mammoth clover.

Natural size.

101 days after planting.

Natural size.

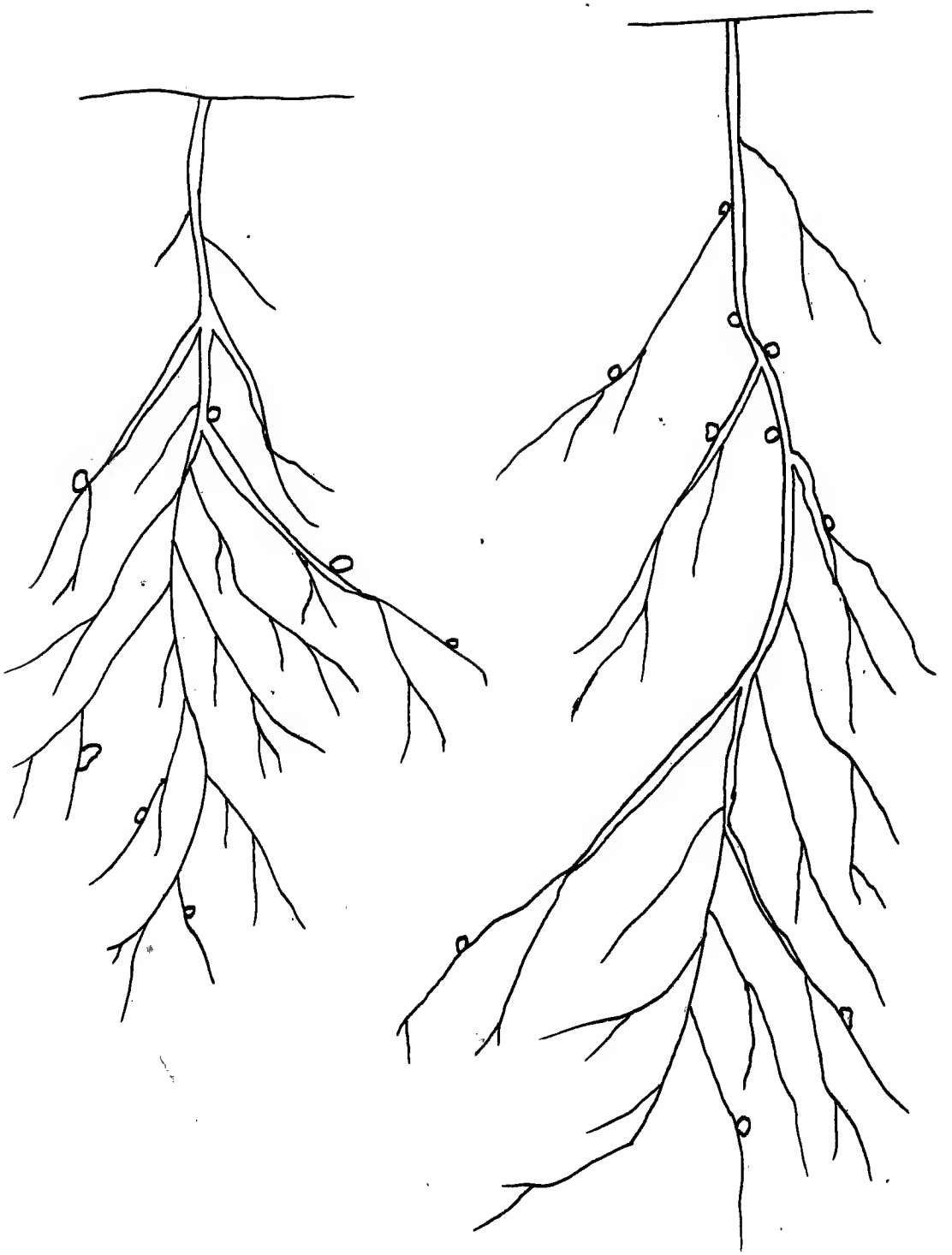
125 days after planting.



Alfalfa (Medicago sativa)

Enlarged twice.
39 days after
planting.

Enlarged twice.
54 days after
planting.



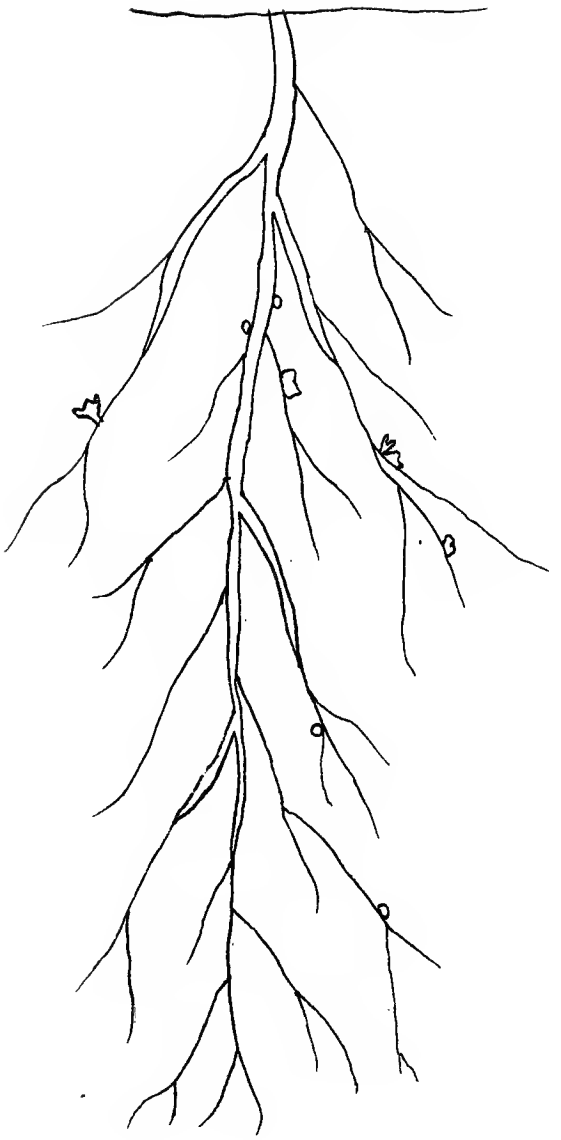
Alfalfa (Medicago sativa)

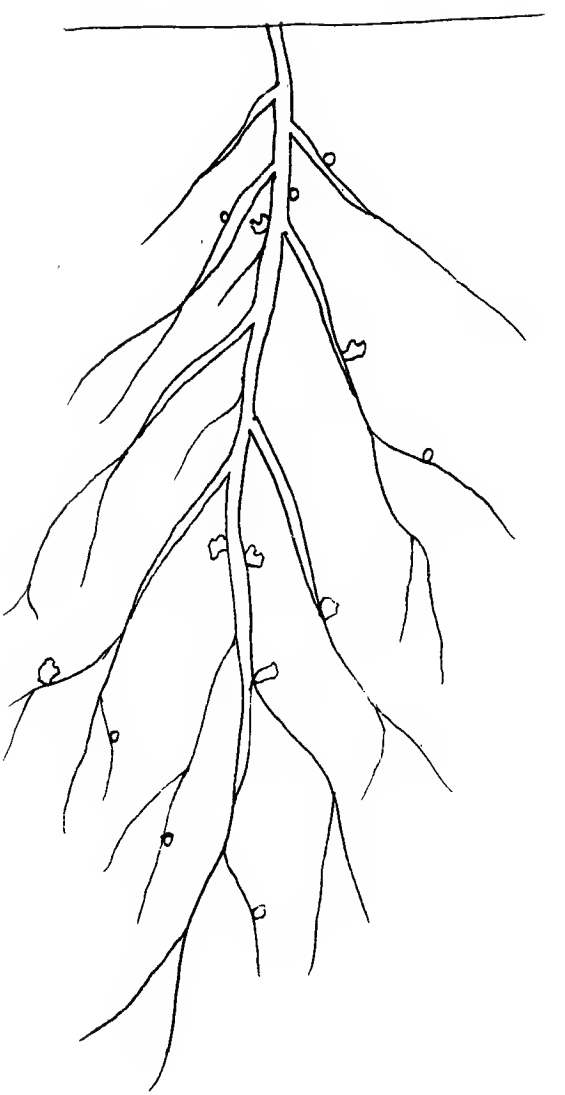
Enlarged twice.

56 days after planting.

Enlarged twice.

86 days after planting.





Alfalfa (mediaago sativa)

Natural size.
98 days after
planting.

Natural size.
108 days after
planting.

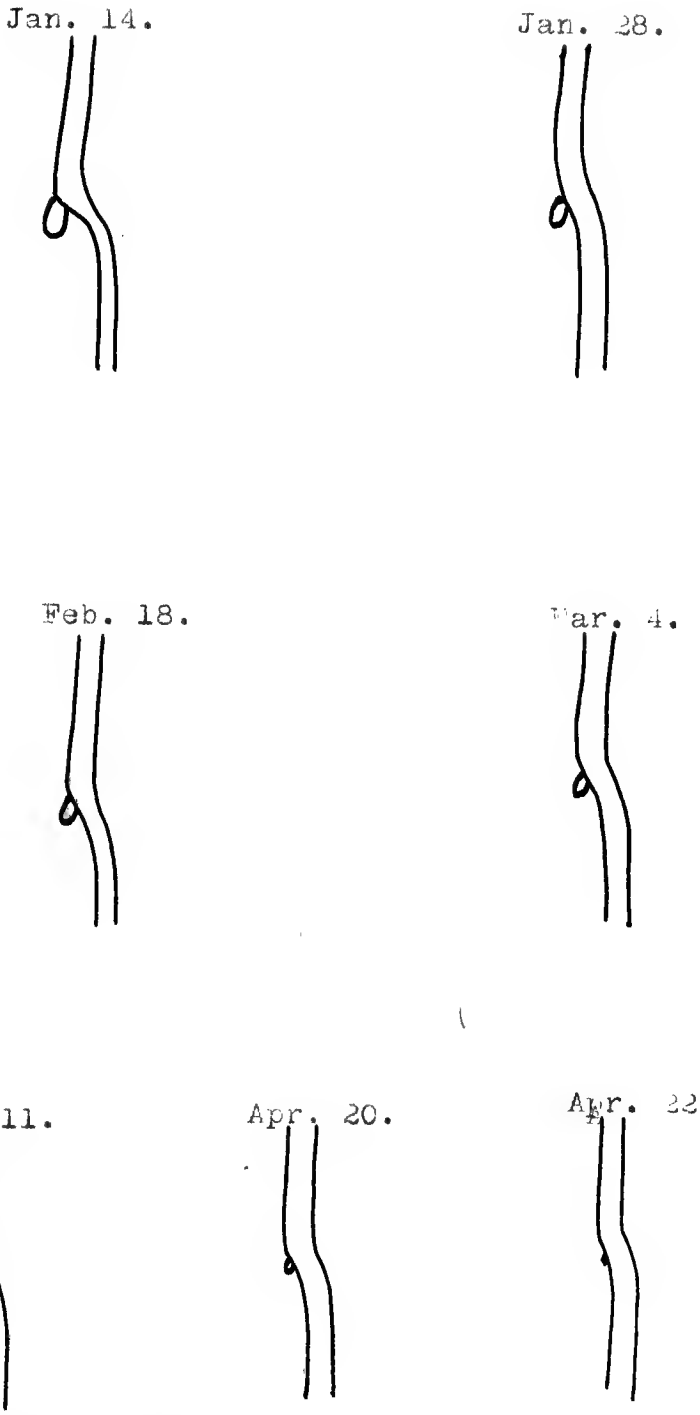


Table showing decrease in the size of nodules.